REMARKS

Claims 9-12, 20-22, 24-26, 28-32, and 34-35 are pending in the application, with claims 9 and 20-22 being currently amended and claims 16-18 and 28 being newly cancelled.

Applicants also submit herewith a certified English translation of the Japanese patent application (See Exhibit A) to which the benefit of foreign priority is claimed from the present application.

Claim 9, which is the only independent claim, has been amended by incorporating therein the subject matter of now cancelled claim 16 to more clearly define over the art of record. In particular, claim 9 now recites a process for producing lactoperoxidase comprising, in part, a step (3) for bringing said washed cation exchanger into contact with a leaching solvent which elutes lactoperoxidase, wherein an ionic strength of the leaching solvent is 0.07 to 0.3, to thereby obtain a leaching solution having lactoperoxidase eluted into said leaching solvent. [underlining for emphasis]. Claim 9 also further recites a step (5) for obtaining a lactoperoxidase solution by removing the precipitation of impurities from said concentrated leaching solution. [underlining for emphasis].

Dependent claims 20-22 have been amended to properly depend, directly or indirectly, from claim 9 in view of the cancellation of claims 16-18.

35 U.S.C. §103 rejections

Previously pending claims 9-12, 16-18, 20-22, 24-26, 28-32, and 34-35 stand rejected under 35 U.S.C. 103(a) as being unpatentable over Uchida U.S. Patent No. 5,596,082 ("Uchida"), Burling U.S. Patent No. 5,149,647 ("Burling"), Kussendrager U.S. Patent Nos. 5,596,082 and 6,010,698 ("the Kussendrager '082 patent" and "the Kussendrager '698 patent", respectively)(collectively, "the Kussendrager patents"), Souppe FR 2841747 as evidenced by U.S.

Patent No. 7,247,331 ("Souppe"), and Lihme U.S. Patent No. 5,780,593 ("Lihme"). *See* Official Action at Pages 3-7. Applicants respectfully disagree with the present rejections, particularly in view of independent claim 9 as currently amended.

By way of background, in the present invention, all proteins are included in a concentrated leaching solution since molecular weight of the proteins is large and the proteins do not pass through an ultrafiltration membrane. On the other hand, in a solution which has been passed through the ultrafiltration membrane, salts or the like which have small molecular weight exist. In the present invention, the concentrated leaching solution obtained in the step 4) of claim 9 has a solution in which lactoperoxidase has been dissolved and precipitate of impurity-proteins exist, since these lactoperoxidase and other proteins do not pass through an ultrafiltration membrane. For example, as shown in Examples of the present specification, ultrafiltration filters each having fractional molecular weight of 10K Dalton, 20K Dalton, 30K Dalton or 50K Dalton are used. Lactoperoxidase which has the molecular weight of about 80K Dalton do not pass through the ultrafiltration membrane.

While the present invention can use an ultrafiltration membrane for concentration and desalting similar to general techniques, the present invention has a superior characteristic in that the ultrafiltration membrane is used to separate lactoperoxidase and other proteins as fractions having different properties. One of the fractions is soluble in a concentrated leaching solution and the other is insoluble therein.

To achieve the aforementioned separation, a treatment wherein a leaching solvent in step 3) having an ionic strength of 0.07 to 0.3 is used for a cation exchange resin, to which absorption of milk materials has been conducted; and a treatment wherein an ultrafiltration membrane in step 4) is used so that the protein content in the leaching solution becomes 0.9 to

15%, are both combined in the present invention. Due to the combination of the treatments, unexpected and excellent effects are achieved such that proteins other than lactoperoxidase are generated as precipitate, i.e., impurities, in a concentrated solution side of an ultrafiltration membrane; and said proteins can be removed easily since said impurity-proteins are insoluble in purified water.

Indeed, the ultrafiltration treatment of the present invention can display not only the effects of desalting and concentrating lactoperoxidase in a lactoperoxidase-containing solution but also the effects of generating precipitate (proteins which are other than lactoperoxidase), which can be easily separated as an impurity in the subsequent step. Further, the solution containing lactoperoxidase, which is obtained after the ultrafiltration treatment, has excellent effects in that it is possible to obtain a solution including high purity lactoperoxidase merely by removing the precipitate of impurities. That is, in the present invention, it is possible to obtain a high purity lactoperoxidase powder, by merely drying the solution including high purity lactoperoxidase from which impurities are removed. In addition, the precipitate of impurities, which has been generated by the ultrafiltration treatment in the lactoperoxidase-containing solution according to the present invention, is no longer dissolved in purified water or the like, even after new purified water or the like is added and concentration is further conducted. Accordingly, the present invention also has an excellent effect in that desalting treatment can be easily conducted. *See* Test Example 2 of the present specification.

Applicants now specifically address the present §103 rejections. Even assuming *arguendo* that one skilled in the art would combine Uchida, Burling, the Kussendrager patents, Souppe, and Lihme, which we assert one would not, the combination still fails to make obvious Applicant's process for producing lactoperoxidase, as now recited in claim 9. Indeed, to establish

prima facie obviousness of a claimed invention, it is certainly well established that all the claim limitations must be taught or suggested by the prior art. *In re Royka*, 490 F.2d 981, 180 USPQ 580 (CCPA 1974); *See also* MPEP §2143.03 (citing *In re Wilson*, 424 F.2d 1382, 1385, 165 USPQ 494, 496 (CCPA 1970))(To establish *prima facie* obviousness of a claimed invention, it is certainly well established that "all words in a claim must be considered when judging the patentability of that claim against the prior art or suggested by the prior art." (emphasis added)). In the instant case, Applicants respectfully submit that the Examiner has failed to establish a *prima facie* case of obviousness for the reasons that follow.

Upon review of Uchida, this reference discloses that, in the manufacturing of lactoperoxidase and the like, lactoperoxidase can be eluted from a cation exchange resin, to which absorption of milk materials has been conducted, with a solution having an ionic strength of from about 0.2 to about 0.5. Then, an ultrafiltration treatment is performed to conduct only desalting and concentration of the leaching solution.

In contrast, the method of the present invention that makes it possible to obtain a high purity lactoperoxidase includes, in part, the combination of a step, i.e., step 3) wherein a cation exchange resin, to which absorption of milk materials has been conducted, is washed with a solution having an ionic strength of from about 0.07 to about 0.3 to elute a solution, which includes lactoperoxidase; and a step, i.e., step 4) wherein the solution to which lactoperoxidase has been eluted is treated by an ultrafiltration treatment so that the protein content in the concentrated solution side of an ultrafiltration membrane becomes 0.9 to 15%. Due to the combination, an insoluble fraction (proteins other than lactoperoxidase) and a soluble fraction (lactoperoxidase) can exist in a concentrated solution side of an ultrafiltration membrane. In a solution that has been passed through the ultrafiltration membrane, neither lactoperoxidase nor

proteins other than lactoperoxidase exist. Furthermore, the insoluble fraction (other proteins) can be simply removed from the concentrated solution to obtain a high purity lactoperoxidase. Such a process is not disclosed or suggested in Uchida.

In other words, due to the treatment using an ultrafiltration membrane, the process for producing lactoperoxidase of the present invention enables unexpected and excellent effects wherein concentration of lactoperoxidase and separation of impurity-proteins are conducted at the same time by the ultrafiltration treatment since a soluble fraction (lactoperoxidase) can be concentrated while other proteins can be separated as an insoluble fraction.

Moreover, the precipitate of other proteins (impurities), which has been generated in a concentrated solution side of the ultrafiltration membrane, is no longer dissolved in purified water even if new purified water is added. Accordingly, the desalting treatment of the lactoperoxidase solution can be easily conducted merely by repeating the concentration treatment wherein purified water is added multiple times. Such an effect of the present invention is not disclosed in Uchida. *See* Test Example 2 of the present specification.

Further concerning Burling, upon review, Burling discloses a microfiltration (MF) to prevent clogging of an ion exchanger, wherein the clogging of an ion exchanger is caused by occurrence of particles of globular fat and protein aggregate in milk materials when cation exchange treatment is conducted at a high rate. Col. 3, lines 44 to 47. Also, the microfiltration is adopted in Burling as a pretreatment for the cation ion exchange treatment. Col. 3, lines 47 to 51. However, the order of production steps in Burling is different from those of the present invention wherein a leaching solution is concentrated with an ultrafiltration membrane after cation exchange treatment is conducted. Furthermore, there is no disclosure at all in Burling that an ultrafiltration method is used for dividing lactoperoxidase as a soluble fraction and impurities as an insoluble

fraction (precipitation). In other words, Burling, like Uchida, simply fails to at least disclose a step for concentrating a leaching solution through an ultrafiltration membrane so that a protein content in the concentrated leaching solution becomes 0.9 to 15% to thereby effect precipitation of impurities in the concentrated leaching solution, as is required by claim 9.

Upon review of the Kussendrager patents, the Kussendrager '082 patent and the Kussendrager '698 patent disclose a process for isolating a lactoferrin solution or growth factors, and also disclose that salts are removed by ultrafiltration. The Kussendrager patents teach a step for a desalting treatment for a lactoferrin-containing fraction and a lactoperoxidase-containing fraction, and disclose that salts are removed as precipitate in said desalting step. However, neither of the Kussendrager patents discloses that a solvent having an ionic strength of 0.07 to 0.3 is used for washing, which is conducted for a cation exchanger to which milk materials have been adsorbed, to obtain a fraction including lactoperoxidase.

Furthermore, neither of the Kussendrager patents discloses that the concentration treatment is conducted so that a protein content in a solution including lactoperoxidase becomes 0.9 to 15%, or discloses effective separation wherein an insoluble fraction (other proteins) and a soluble fraction (lactoperoxidase) are generated in a concentrated solution side of an ultrafiltration membrane due to the combination of the above concentration treatment and eluting-conditions for the cation exchanger. That is, the present invention does not use an ultrafiltration membrane only for the concentration and desalting. Rather, an ultrafiltration membrane is also used for separating a mixture of lactoperoxidase and other proteins in milk materials into an insoluble fraction (other proteins) and a soluble fraction (lactoperoxidase) wherein the fractions have different properties.

Again, by combining a treatment wherein a leaching solvent having an ionic strength of 0.07 to 0.3 is used and a treatment wherein an ultrafiltration membrane is used so that a protein content in the leaching solution becomes 0.9 to 15%, unexpected and excellent effects of can be obtained wherein impurities (proteins other than lactoperoxidase), which are generated at the concentrated solution side of an ultrafiltration membrane, can be separated as a precipitate that does not dissolve in purified water. As a result, it is easy to remove the precipitate having such a property. In this way, the present invention has unexpected superior effects.

Upon review of Souppe, Souppe discloses a method wherein milk materials are adsorbed by a cation exchange resin, then the adsorbed milk materials are eluted with a salt solution, and desalting is carried out by an ultrafiltration method. Col. 2, lines 44 to 54. However, there is no disclosure at all in Souppe that an ultrafiltration method is used for dividing lactoperoxidase as a soluble fraction and impurities as an insoluble fraction (precipitation). In other words, Souppe, like Uchida, Burling, and the Kussendrager patents, simply fails to at least disclose or suggest a step for concentrating a leaching solution through an ultrafiltration membrane so that a protein content in the concentrated leaching solution becomes 0.9 to 15% to thereby effect precipitation of impurities in the concentrated leaching solution, as is required by claim 9.

Finally, upon review of Lihme, Lihme discloses an ultrafiltration method which is conducted for concentration, desalting and the like. Col. 6, lines 12 to 18. However, there is disclosure in Lihme that an ultrafiltration method is used for dividing lactoperoxidase as a soluble fraction and impurities as an insoluble fraction (precipitation). In other words, Lihme, like the Uchida, Burling, the Kussendrager patents, and Souppe, simply fails to at least disclose a step for concentrating a leaching solution through an ultrafiltration membrane so that a protein content in

the concentrated leaching solution becomes 0.9 to 15% to thereby effect precipitation of impurities in the concentrated leaching solution, as is required by claim 9.

In view of the above, when combined, Uchida, Burling, the Kussendrager patents, Souppe, and Lihme fail to provide all of the elements of Applicants' claimed process for producing lactoperoxidase. That is, Examiner has not established a *prima facie* case of obviousness based on these disclosures insofar as these references, collectively, fail to at least disclose the combination of a treatment wherein a leaching solvent in step 3) having an ionic strength of 0.07 to 0.3 is used for a cation exchange resin, to which absorption of milk materials has been conducted; and a treatment wherein an ultrafiltration membrane in step 4) is used so that the protein content in the leaching solution becomes 0.9 to 15%.

Further, as is apparent from the results of Test Example 2 of the present specification, the present invention has unexpected results with excellent effects, wherein even if purified water or the like is added to the concentrated fraction obtained by the ultrafiltration treatment, and subsequent concentration is further conducted again, the precipitation is no longer dissolved in the purified water or the like. That is, the present invention can have synergistic effects wherein both treatments of removing impurities and treatments of demineralization and concentration can be conducted simultaneously. Accordingly, it would not be within the purview of a skilled artisan to expect the specific and excellent effects of the present invention from the cited references.

For all of the above reasons, the rejections are overcome and must be withdrawn. Applicants, thus, respectfully submit that independent claim 9, along with its dependent claims, is allowable over the cited references.

Conclusion

As a result of the remarks given herein, Applicants submit that the rejection of the pending claims has been overcome. Therefore, Applicants respectfully submit that this case is in condition for allowance and request allowance of the pending claims.

If Examiner believes any detailed language of the claims requires further discussion, Examiner is respectfully asked to telephone the undersigned attorney so that the matter may be promptly resolved. Applicants also have submitted all fees believed to be necessary herewith. Should any additional fees or surcharges be deemed necessary, Examiner has authorization to charge fees or credit any overpayment to Deposit Account No. 23-3000.

Respectfully submitted,
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